PATHOLOGY AND BACTERIOLOGY.

UNDER THE CHARGE OF
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IDENTIFICATION OF THE SPIRILLUM OF ASIATIC CHOLERA.

In the course of an exhaustive study of the cholera vibrio, more particularly as regards the artificial induction of immunity to its inoculation in animals, R. Pfeiffer discovered the interesting fact that when mixed with the serum of immunized guinea-pigs, and inoculated into the peritoneal cavity of susceptible animals, virulent cultures of the spirilla in large dose remained innocuous, and that on subsequent examination of the peritoneal contents the bacteria could be seen to have undergone disintegration to a greater or less extent, dependent upon the relative immunizing strength of the serum of the immunized animal. This power of destroying the cholera spirilla is believed by Pfeiffer to depend upon the presence in the serum of certain antagonistic substances ("Antikörper") which have a distinct inhibiting influence upon the vital processes of the bacteria. As a result of this discovery PFEIFFER (*Zeitschrift f. Hygiene und Infectionskrankheiten*, 1895, xix. p. 75) has suggested a test of the identity of the cholera spirillum, for his investigations show that no other species of bacteria is affected in the same way by mixing with the serum. It will be remembered that a similar test was alluded to in connection with the identification of the typhoid bacillus (*Cf.* AMERICAN JOURNAL OF THE MEDICAL SCIENCES, May, 1896, p. 624).

The method suggested by Pfeiffer is as follows: A loopful of the culture to be tested is mixed with a cubic centimetre of bouillon, to which ten times the amount of serum necessary to protect a guinea-pig of 200 grammes weight from a similar dose of virulent cholera spirilla has been added, and the whole is at once inoculated into the peritoneal cavity of a young guinea-pig of from 200 to 300 grammes weight. In the inoculation care should be taken to avoid injury of the intestines, and the cultures employed should be recent and should have been shown to consist of well-formed and actively moving germs.

As control, a similar quantity of the same culture is mixed with a cubic centimetre of bouillon as before, an amount of ordinary guinea-pig serum equal to the amount of immunizing serum made use of in the original test is added, and the whole is inoculated into another guinea-pig.

In twenty minutes some of the peritoneal contents in each case is withdrawn by means of glass pipettes, and is examined. If the bacteria are the specific germs of cholera they present a very different appearance in the two cases. Those obtained from the control-animal are well formed, active, and seem to have multiplied. Those which were exposed to the action of the immunizing serum are small, misshapen, immobile for the most part, apparently dead. Pfeiffer maintains that unless a distinct difference is observable between the bacteria in the two experiments the micro-organism under examination must be regarded as probably *not* the cholera vibrio, since the change described is very constant in the case of the cholera germ, and has not been observed to occur with any other under similar conditions. Pfeiffer accordingly presents this as a specific differential test for the cholera bacillus.

Since the publication of the above test a simpler modification of it, not requiring the inoculation of animals, has been suggested by PFEIFFER and VAGEDES (*Centralblatt f. Bakteriologie*, etc., 1896, xix. No. 11, 385), and has been found to be of great use in distinguishing the pathogenic species from other closely allied forms.

In the course of experiments directed to the explanation of the disintegration of the bacteria which occurs when they are mixed with immunizing serum and inoculated, it was discovered by Pfeiffer that the bacilli did not undergo disintegration when they were exposed to the action of the serum in a test-tube only. In this case the activity of the germs was inhibited, and they collected at the bottom of the test-tube in flocculi, but their shape remained unaltered. It was further noted that after a variable time in the incubator the flocculi began to break up, microscopic examination showing that the germs were recovering their mobility and were gradually returning to their normal condition. From this Pfeiffer inferred that the effect of the immunizing serum is to inhibit temporarily the activity of the germs, but that without the body-juices it is incapable of destroying them.

The test evolved from these observations depends upon the fact that no other germ than the cholera-bacillus is similarly inhibited in its activity by the specific immunizing serum. As suggested by Pfeiffer and Vagedes, it is carried out in the "hanging drop." The serum employed by them had an immunizing power such that one-fifteenth milligramme was capable of protecting a 200-gramme guinea-pig against a dose of 2 milligrammes of virulent cholera-culture. This serum was diluted with fifty times its quantity of bouillon, and hanging drops of it were inoculated with the culture to be tested. The microscope showed the gradual inhibition of the activity of the cholera-bacilli, resulting in the complete inactivity of all present after twenty minutes. The cultures were then placed in the incubator and examined from time to time. In the course of twenty-four hours the activity had returned, and the germs had undergone considerable multiplication.

The constancy of this test may be inferred from the fact that Pfeiffer and Vagedes tested seventy different cultures of the cholera-bacillus and twenty of various other closely allied species, and found it reliable in all. The cholera-germs were always inhibited in their activity; the other species, never.

Pfeiffer announces that he is prepared to furnish serum of a sufficient immunizing strength for this test to any who desire to confirm the reliability of the test or to determine the nature of any suspected cultures.

THE CAUSE OF DEATH IN ACUTE LOBAR PNEUMONIA.

Some recent observations of BOLLINGER (*Münchener med. Wochenschrift*, 1895, No. 32) made at autopsies in cases of pneumonia are of interest as